

What is claimed is:

1. A method for screening a test substance for COX-2 inhibitory activity comprising:

(a) contacting the test substance with indicator cells which constitutively express endogenous COX-2 or inducibly express endogenous COX-2; and

(b) determining the level of:

(i) proliferation of the indicator cells in the presence and absence of the test substance, a decreased level of proliferation of the indicator cells in the presence of the test substance indicating that the test substance has COX-2 inhibitory activity; or

(ii) one or more prostaglandins produced by the indicator cells in the presence and absence of the test substance, a decreased prostaglandin level in the presence of the test substance indicating that the test substance has COX-2 inhibitory activity.

(iii) arachidonic acid produced by the indicator cells in the presence and absence of the test substance, an increased arachidonic acid level in the presence of the test substance indicating that the test substance has COX-2 inhibitory activity.

2. A method for screening a test substance for COX-2 inhibitory activity comprising:

(a) contacting the test substance with indicator cells which express a GTPase-deficient mutant form of the α -subunit of protein G12, which mutant α -subunit has the capacity to induce the production of arachidonic acid and COX-2 in the indicator cells; and

(b) determining the level of proliferation of the indicator cells in the presence and absence of the test substance, a decreased level of proliferation of the indicator cells in the presence of the test substance indicating that the test substance has COX-2 inhibitory activity.

3. A method according to claim 2 wherein the G12 protein α -subunit mutant comprises the Q229L mutation.

4. A method according to claim 3 wherein the level of indicator cell proliferation is determined by an assay for DNA synthesis by the indicator cells.

5. A method according to claim 4 wherein the DNA synthesis assay comprises assaying tritium-labeled thymidine uptake by the indicator cells.

6. A method for screening a test substance for COX-2 inhibitory activity comprising:

(a) contacting the test substance with indicator cells which express a GTPase-deficient mutant form of the α -subunit of protein G12, which mutant α -subunit has the capacity to induce the production of arachidonic acid and COX-2 in the indicator cells; and

(b) determining the level of one or more prostaglandins produced by the indicator cells in the presence and absence of the test substance, a decrease in production of said prostaglandin by the indicator cells in the presence of the test substance indicating that the test substance has COX-2 inhibitory activity.

7. A method according to claim 6 wherein the G12 protein α -subunit mutant comprises the Q229L mutation.

8. A method according to claim 7 wherein the prostaglandin level is assayed in the media surrounding the indicator cells, in the indicator cells or in a component of the indicator cells.

9. A method according to claim 8 wherein the prostaglandin level

is assayed by a prostaglandin immunoassay.

10. A method according to claim 9 wherein the prostaglandin immunoassay comprises a competitive immunoassay.

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~~12.~~ A method for screening a test substance for COX-2 inhibitory activity comprising:

(a) contacting the test substance with indicator cells which express a GTPase-deficient mutant form of the α -subunit of protein G12, which mutant α -subunit has the capacity to induce the production of arachidonic acid and COX-2 in the indicator cells; and

(b) determining the level of arachidonic acid provided by the indicator cells in the presence and absence of the test substance, an increase in the level of arachidonic acid provided by the indicator cells in the presence of the test substance indicating that the test substance has COX-2 inhibitory activity.

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~~13.~~ A method according to claim 12 wherein the G12 protein α -subunit mutant comprises the Q229L mutation.

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~~14.~~ A method according to claim 13 wherein the arachidonic acid level is assayed in the media surrounding the indicator cells, in the indicator cells or in a component of the indicator cells.

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~~15.~~ A method according to claim 14 wherein the arachidonic acid assay comprises assaying tritium labeled arachidonic acid release from said indicator cells.

CELL-BASED ASSAY FOR SCREENING COX-2 INHIBITORS

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